

John Harada, *et al.*
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28-29, 35-36, 39, 42-43, 47-49, 54-55, 58, 63, 69, and 70-73 are pending and under consideration with entry of this Amendment.

A marked up copy of amended claims 1, 21, 35, 42, 47, 54, and 55 are provided as Appendix A entitled "**VERSION WITH MARKINGS TO SHOW CHANGES MADE.**" As a convenience to the Examiner, a complete set of the claims, as amended herein, is also attached to this Amendment as Appendix B.

2. *Support for the Amendments*

Support for the amendments to the claims can be found throughout the specification, the drawings, and the claims as originally drafted. For example, support for "modulation of embryo development in plants expressing LEC1 polynucleotides or polypeptides can be found, e.g., on page 12, lines 12-16 of the specification. Support for claims 70-73 can be found, e.g., on page 18, lines 10-15 of the present specification. No new matter is added.

3. *Interview*

Applicants thank the Examiner for the helpful interview.

4. *Claim objections*

Claims 35 and 42 were objected to for depending from non-elected claims. As amended, claims 35 and 42 do not depend from non-elected claims. Therefore, withdrawal of the rejection is requested.

Claim 54 was objected to for misspelling "embryonic." Claim 55 was objected to for lacking a period. Applicants thank the Examiner for noting these typographical errors. The errors are corrected in the present Amendment.

5. *Rejections under 35 U.S.C. § 112, first paragraph*

Claims 21, 22, 28, 29, and 39 were rejected under the written description requirement of section 112, first paragraph. In addition, claims 1-3, 9, 21-22, 28-29, 35-

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36, 39, 42-43, 47-49, 54-55, 58, 63, and 69 were rejected as allegedly not enabled by the specification. According to the Examiner, this rejection is based on the scope of polynucleotides encompassed by the claims. Applicants respectfully traverse the rejection.

A. Written Description Rejection

Applicants submit that the Federal Circuit has held that the written description requirement can be fulfilled in any number of ways, so long as the specification describes the invention "in sufficient detail that one skilled in the art can clearly conclude that the inventor invented the claimed invention." *See University of California v. Eli Lilly & Co.*, 43 USPQ2d 1398, 1404 (Fed. Cir. 1997). For a chemical invention, an adequate description "requires a precise definition, such as by **structure**, formula, chemical name, or **physical properties**...." (Emphasis added). Accordingly, as described below, the present specification provides ample written description for the pending claims, precisely as required by the Court in *University of California*.

In the present case, the claims are directed polynucleotides that encode polypeptides at least 80% identity to the B domain of SEQ ID NO:2, wherein the polypeptides modulate embryo development when expressed in plants. This claim language defines a *physical* and *structural property* of the invention, as explicitly required by the court in *University of California*. Percent identity to a particular sequence reflects the *structure* of the nucleic acid, *i.e.* that its primary *structure*, or nucleotide sequence, is similar to the recited sequence. Thus, the description of the claimed invention satisfies the written description requirement as set forth by the court in *University of California* on at least two grounds, *i.e.* structure and physical properties.

B. Enablement

It is Applicant's understanding that the Examiner's concerns center on the ability of polypeptides within the scope of the claims to function as described in the specification. Applicants submit that specification provides methods of constructing and

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testing polynucleotide of the invention for function. The application also provides a working example demonstrating that the L1L protein, which has approximately 80% identity with LEC1 (SEQ ID NO:2), can act to complement a *lec1* mutation. This example demonstrates that sequences encoding a polypeptide at least 80% identical to SEQ ID NO:2 can function to modulate embryo development. In addition, Applicants have provided a declaration of John J. Harada, Ph.D., illustrating the relationship of LEC1 and L1L in the B domain as well as additional evidence demonstrating that polypeptides comprising a subsequence about 68% (i.e., lower than the 80% claimed) identical to the B domain in SEQ ID NO:2 are functional. Therefore, Applicants respectfully submit that the claims meet all requirements under 35 U.S.C. § 112.

i. The specification describes structural guidelines for the function of LEC1 polypeptides in plants

The present specification describes the Arabidopsis LEC1 polypeptide, other related sequences and their effect of embryo development. For example, the specification demonstrates that plants with mutations in LEC1 have defects in late embryogenesis and that this phenotype can be complemented upon introduction of polynucleotides encoding SEQ ID NO:2 into the mutants. *See, e.g.,* page 33, line 24 to page 34, line 23 and page 35, lines 20-32. In addition, the specification notes LEC1's structural relation to the HAP3 and HAP3 homologs, including homology within the B domain. *See, e.g.,* page 18, line 28 to page 19, line 22 of the specification. Thus, Applicants asserted that polypeptides at least 80% identical to the B domain modulate embryo development. *See, e.g.,* page 18, lines 10-16 (describing the B domain) and page 10, lines 22-25, describing polypeptides at least 80% identical to a reference sequence (e.g., the B domain).

Those of ordinary skill in the art were capable of identifying functionally active LEC1 polypeptides within the scope of the claims by employing the methods described in the specification. For example, the application describes various phenotypes associated with modulated LEC1 expression. Thus, candidate sequences can be

introduced into plants and functional sequences can be identified based on their ability to modulate embryo development. Ectopic expression can be used to induce expression of embryonic characteristics such as the accumulation of seed storage proteins in non-seed tissues. *See, e.g.*, page 17, lines 19-21 of the specification. Inhibition of endogenous LEC1 expression in plants results in, *e.g.*, plants with non-viable seed. *See, e.g.*, page 15, lines 1-2 of the specification. Moreover, the specification teaches how to construct polynucleotides of the invention and transform them into plants. Therefore, those of ordinary skill in the art could have readily identified functional polypeptides within the scope of the claims.

ii. *L1L exemplifies a polypeptide with approximately 80% identity to SEQ ID NO:2*

The L1L polypeptide provides further evidence that polypeptides at least 80% identical to SEQ ID NO:2 function to modulate embryo development. As described on page 41-42 of the present specification, L1L complements *lec1* mutant plants. Whereas progeny of *lec1* mutant plants die following desiccation, mutants transformed with L1L under the control of the LEC1 promoter produced viable seed that resembled wild type plants. *See, e.g.*, page 41, lines 29-32 of the present specification.

The Harada Declaration, which accompanies this Amendment, presents an alignment of LEC1 and L1L. LEC1 and L1L are 83.3% identical within the B domain (amino acids 27-117 of SEQ ID NO:2). *See*, Harada Declaration, paragraph 7 and Figure 3. In contrast, the A and C domains of LEC1 and L1L have almost no similarity. The Harada Declaration also demonstrates that the L1L polypeptide modulates embryo development when introduced into plants. *See*, Harada Declaration, paragraph 7. These results demonstrate that polypeptides comprising subsequences about 80% identical to the B domain in SEQ ID NO:2 are active and modulate embryo development in plants. Therefore, the results described in the specification demonstrate that polypeptides comprising a sequence at least 80% identical to the B domain of SEQ ID NO:2 modulate embryo development in plants.

iii. *The Harada Declaration demonstrates that sequences with even less than 80% identity to the B domain of SEQ ID NO:2 modulate embryo development*

The Harada Declaration also demonstrates that a polypeptide with only 68% sequence identity to the B domain of SEQ ID NO:2 can modulate embryo development in plants. The Harada Declaration states that a single amino acid at position 28 in the B domain of At4g14540 was changed from lysine (K) to aspartic acid (D). The aspartic acid residue is present at the corresponding position in LEC1 and L1L but not in other Arabidopsis HAP3 homologs. The resulting protein, "K28D At4g14540," has 67.2% identity to SEQ ID NO:2 within the B domain and displayed significant activity in an assay measuring LEC1 function. *See*, Harada Declaration, paragraph 8 and Figure 4.

These results demonstrate that the present claims (directed to polypeptides comprising sequences at least 80% identical to the B domain of SEQ ID NO:2) are well within the scope of active variants of the LEC1 sequence. Since sequences at least 68% identical to the B domain of SEQ ID NO:2 are active, there is no reason why sequences with 80% identity would not work. Thus, the Harada Declaration further confirms that those of skill in the art could have used routine methods to generate active sequences within the scope of the claims.

iv. *Conclusion*

The specification provides methods of making and identifying functional polypeptides of the invention, i.e., polypeptides that modulate embryo development in plants. Moreover, the application demonstrates that polypeptides comprising an amino acid sequence at least about 80% identical to the B domain of SEQ ID NO:2 modulate embryo development. Indeed, the results described in the specification demonstrate that such polypeptides can complement mutations in SEQ ID NO:2. The Harada Declaration confirms these results and provides further evidence that sequences with even less identity than is claimed are active. In light of the data present in the specification, as well

as the information provided in the Harada Declaration, Applicants respectfully submit that the amended claims fulfill all of the requirements of 35 U.S.C. § 112.

6. *Rejections under 35 U.S.C. § 112, second paragraph*

Claim 21 was rejected as allegedly clear for reciting the term "clone MNJ7." As amended, claim 21 recites the Genbank accession number for the clone, thereby eliminating any possible ambiguity in the term. Accordingly, Applicants request withdrawal of the rejection.

Claims 47, 54 and 55 were rejected as allegedly unclear in reciting "modulated" and "modulating." Applicants submit that the term "modulated" is a common term meaning "changed" or "altered". For example, as amended, claim 47 provides a method of modulating embryo development in a plant. The method provides for induction of ectopic embryo development by ectopic expression of the polypeptides of the invention. The methods also provides for preventing embryo development in tissues where LEC1 polypeptides are expressed endogenously. Thus, in light of the specification and the common meaning of the term, Applicants submit that the term "modulated" is clear. Accordingly, withdrawal of the rejection is respectfully requested.

7. *Rejections under 35 U.S.C. § 102*

Claims 21, 22, 28 and 29 were rejected under 35 U.S.C. § 102(b) as allegedly anticipated by Genbank accessions submitted by Lotan *et al.* and Feng *et al.* that became publicly available on July 2, 1998 and October 7, 1998, respectively. In addition, claims 1-3, 9, 21-22, 28-29, 35-36, 39, 42-43, 47-49, 54-55, 58, 63 and 69 were rejected as allegedly anticipated by a *Cell* paper by Lotan *et al.*, published June 26, 1998. Applicants respectfully traverse the rejections.

The present specification provides the following priority listing on page 1:

The present application is a Continuation-In-Part ("CIP") of United States Patent Application Serial Number (USSN) 09/193,931, filed November 17, 1998, which

is a CIP of USSN 09/103,478, filed June 24, 1998, which is a CIP of USSN 09/026,221, filed February 19, 1998, which is a CIP of USSN 08/804,534, filed February 21, 1997.

The claimed subject matter claims priority to at least USSN 09/103,478, filed June 24, 1998, now U.S. Patent No. 6,235,975, issued May 22, 2001. For example, USSN 09/103,478 describes the presence of the B domain in the LEC1 sequence and states that LEC1 polypeptides share a high homology with the B domains of the yeast HAP3 protein. *See, e.g.*, page 37, lines 1-13 of USSN 09/103,478. Eighty percent sequence identity finds support on, *e.g.*, page 14, line 28 of USSN 09/103,478. Since the claims at issue have priority to a filing date before any of the cited references were publicly available, the references cannot anticipate the present claims. Accordingly, Applicants respectfully request withdrawal of the rejections.

8. Double Patenting Rejection

Claims 1-3, 9, 21-22, 28-29, 35-36, 39, 42-43, 47-49, 54-55, 58, 63 and 69 were rejected for alleged obviousness-type double patenting. Applicants will gladly consider providing the Examiner with a terminal disclaimer after the Examiner has indicated that claimed subject matter is otherwise allowable.

CONCLUSION

In view of the foregoing, Applicants believe all claims now pending in this Application are in condition for allowance. The issuance of a formal Notice of Allowance at an early date is respectfully requested.

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PATENT

If the Examiner believes a telephone conference would expedite prosecution of this application, please telephone the undersigned at 415-576-0200.

Respectfully submitted,



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APPENDIX A

VERSION WITH MARKINGS TO SHOW CHANGES MADE

1. (Amended) An expression cassette comprising a promoter operably linked to a heterologous polynucleotide sequence, or a complement thereof, encoding a LEC1 polypeptide, comprising a subsequence at least [68%] 80% identical to the B domain of SEQ ID NO:2, wherein the polynucleotide sequence is heterologous to any element in the expression cassette, and wherein the polynucleotide modulates embryo development when the polynucleotide is expressed in a plant.

21. (Amended) An isolated nucleic acid or complement thereof, encoding a LEC1 polypeptide comprising a subsequence at least [68%] 80% identical to the B domain of SEQ ID NO:2, with the proviso that the nucleic acid is not clone MNJ7 (Genbank Accession No. AB025628), wherein the LEC1 polypeptide modulates embryo development when expressed in a plant.

35. (Amended) A host cell comprising an expression cassette according to any of claim [claims] 1[, 15 and 17] or a nucleic acid molecule according to claim 21, wherein the expression cassette or nucleic acid molecule is flanked by a heterologous sequence.

42. (Amended) A method of introducing an isolated nucleic acid into a host cell comprising:

- (a) providing an expression cassette according to any of claim [claims] 1[, 15 and 17] or an isolated nucleic acid according to claim 21; and
- (b) contacting the expression cassette or nucleic acid with the host cell under conditions that permit insertion of the nucleic acid into the host cell.

47. (Amended) A method of modulating embryo development in a plant [transcription], the method comprising,
introducing into the plant an expression cassette containing a plant promoter operably linked to a heterologous LEC1 polynucleotide, the heterologous LEC1 polynucleotide encoding a LEC1 polypeptide comprising a subsequence at least [68%] 80% identical to the B domain of SEQ ID NO:2; and
detecting a plant with modulated embryo development [transcription].

54. (Amended) The method of claim 47, wherein modulating transcription results in the induction of embryonic [embyonic] characteristics in a plant.

55. (Amended) The method of claim 47, wherein modulating transcription results in the induction of seed development.

APPENDIX B

**CLAIMS PENDING AND UNDER CONSIDERATION UPON ENTRY OF
AMENDMENT**

1. (Amended) An expression cassette comprising a promoter operably linked to a heterologous polynucleotide sequence, or a complement thereof, encoding a LEC1 polypeptide, comprising a subsequence at least 80% identical to the B domain of SEQ ID NO:2, wherein the polynucleotide sequence is heterologous to any element in the expression cassette, and wherein the polynucleotide modulates embryo development when the polynucleotide is expressed in a plant.
2. The expression cassette of claim 1, wherein the B domain comprises a polypeptide sequence between about amino acid residue 28 and about residue 117 of SEQ ID NO:2.
3. The expression cassette of claim 1, wherein the B domain comprises a polypeptide sequence with an amino terminus at amino acid residues 28-35 and a carboxy terminus at amino acid residues 103-117 of SEQ ID NO:2.
9. The expression cassette of claim 1, wherein the promoter is a constitutive promoter.
21. (Amended) An isolated nucleic acid or complement thereof, encoding a LEC1 polypeptide comprising a subsequence at least 80% identical to the B domain of SEQ ID NO:2, with the proviso that the nucleic acid is not clone MNJ7 (Genbank Accession No. AB025628), wherein the LEC1 polypeptide modulates embryo development when expressed in a plant.

22. The isolated nucleic acid of claim 21, wherein the B domain comprises a polypeptide sequence with an amino terminus at amino acids 28-35 and a carboxy terminus at amino acids 103-117 of SEQ ID NO:2.

28. The isolated nucleic acid of claim 21, wherein the nucleic acid further comprises a promoter operably linked to the LEC1-encoding nucleic acid.

29. The isolated nucleic acid of claim 29, wherein the promoter is a constitutive promoter.

35. (Amended) A host cell comprising an expression cassette according to any of claim 1 or a nucleic acid molecule according to claim 21, wherein the expression cassette or nucleic acid molecule is flanked by a heterologous sequence.

36. The host cell of claim 35, comprising an expression cassette of claim 1.

39. The host cell of claim 35, comprising a nucleic acid molecule of claim 21.

42. (Amended) A method of introducing an isolated nucleic acid into a host cell comprising:

- (a) providing an expression cassette according to any of claim 1 or an isolated nucleic acid according to claim 21; and
- (b) contacting the expression cassette or nucleic acid with the host cell under conditions that permit insertion of the nucleic acid into the host cell.

43. The method of claim 42, providing the expression cassette of claim 1.

46. The method of claim 42, providing the nucleic acid of claim 21.

47. (Amended) A method of modulating embryo development in a plant, the method comprising,

introducing into the plant an expression cassette containing a plant promoter operably linked to a heterologous LEC1 polynucleotide, the heterologous LEC1 polynucleotide encoding a LEC1 polypeptide comprising a subsequence at least 80% identical to the B domain of SEQ ID NO:2; and

detecting a plant with modulated embryo development.

48. The method of claim 47, wherein the LEC1 polynucleotide encodes SEQ ID NO:2.

49. The method of claim 48, wherein the LEC1 polynucleotide is SEQ ID NO:1.

54. (Amended) The method of claim 47, wherein modulating transcription results in the induction of embryonic characteristics in a plant.

55. (Amended) The method of claim 47, wherein modulating transcription results in the induction of seed development.

58. A transgenic plant cell or transgenic plant comprising the recombinant expression cassette of claim 1.

63. The transgenic plant cell or transgenic plant of claim 58, wherein the promoter is a constitutive promoter.

69. A plant which has been regenerated from a plant cell according to 58.

70. (New) The expression cassette of claim 1, wherein the B domain comprises a polypeptide sequence between amino acid residue 28 and residue 117 of SEQ ID NO:2.

71. (New) The isolated nucleic acid of claim 21, wherein the B domain comprises a polypeptide sequence between amino acid residue 28 and residue 117 of SEQ ID NO:2.

72. (New) The host cell of claim 35, wherein the B domain comprises a polypeptide sequence between amino acid residue 28 and residue 117 of SEQ ID NO:2.

73. (New) The method of claim 47, wherein the B domain comprises a polypeptide sequence between amino acid residue 28 and residue 117 of SEQ ID NO:2.

CURRICULUM VITAE

JOHN JIRO HARADA

I. BIOGRAPHICAL DATA

A. ADDRESS

1. UNIVERSITY: Section of Plant Biology
Division of Biological Sciences
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E-mail: jjharada@ucdavis.edu
2. HOME: 115 Grande Avenue
Davis, CA 95616
telephone: (530) 756-7042

B. BIRTHDATE: March 7, 1953

C. CITIZENSHIP: United States of America

D. MARTIAL STATUS: Married, with three children

II. EDUCATION

A. UNDERGRADUATE

1. DEGREE: B.S., Biochemistry, 1975
2. INSTITUTION: University of California, Los Angeles
3. DATES OF ATTENDANCE: October, 1970 - March, 1975
4. RESEARCH EXPERIENCE: Undergraduate research on plant diterpene metabolism in the laboratory of Dr. Charles A. West, Department of Chemistry and Biochemistry

B. GRADUATE

1. DEGREE: Ph.D., Biochemistry, 1981
2. INSTITUTION: University of Washington, Seattle
3. DATES OF ATTENDANCE: September, 1975 - December, 1980
4. THESIS ADVISOR: Dr. David R. Morris, Department of Biochemistry
5. PH.D. DISSERTATION: Proliferation of Polyamine-Limited Chinese Hamster Ovary Cells

III. PROFESSIONAL EXPERIENCE

- A. **POSTGRADUATE RESEARCH ASSOCIATE**
Laboratory of Dr. Robert B. Goldberg
Department of Biology
University of California, Los Angeles
Los Angeles, CA 90024
February, 1981 - August, 1984
- B. **ASSISTANT PROFESSOR**
Department of Botany
University of California, Davis
Davis, CA 95616
September, 1984 - June, 1990
- C. **ASSOCIATE PROFESSOR**
Department of Botany
University of California, Davis
Davis, CA 95616
July, 1990 - June, 1994
- C. **PROFESSOR**
Section of Plant Biology
Division of Biological Sciences
University of California, Davis
Davis, CA 95616
July, 1994 - present

IV. HONORS

President's Fellowship, University of California, 1970-71

California State Scholarship, 1970-74

Annual Fund Doctoral Fellowship, University of Washington, 1977-78

Faculty Development Award, University of California, Davis, 1988-89

V. PROFESSIONAL ACTIVITIES

A. MEMBERSHIP IN SOCIETIES

American Association for the Advancement of Science

American Society of Plant Physiologists

International Society for Plant Molecular Biology

University of California, Berkeley

B. SERVICE ON REVIEW PANELS

Ad Hoc Panel Member, NIH Biological Sciences Study Section, March, 1991

Member, Committee of Visitors to Review the NSF Developmental Biology Program, July, 1991

Member, Advisory Panel for the NSF Developmental Mechanisms Cluster, April, 1992 - March, 1997

Member, Integrative Graduate Education and Research Training Program, September, 1998

NSERC Site Visit Committee, McGill University, January, 2000

Member, Advisory Panel for the NSF Living Stocks Collection Program, December 2000

Member, Review Panel for the DOE Energy Biosciences Program, December 2001

C. SERVICE AS REVIEWER

1. EDITORIAL BOARDS

Guest Editor, *Plant Molecular Biology*, March, 1992 - March, 1997

Advisory Board, *Plant Physiology and Biochemistry*, March, 1994 - January, 2000

Co-Editor, *The Plant Cell*, July, 1996 - present

Advisory Board, *Journal of Plant Biology*, March, 1998 - present

Editorial Board, *Plant and Cell Physiology*, January 1999 - present

2. JOURNALS

Archives of Biochemistry and Biophysics

Biochemistry

Development

Gene

Genetics

Journal of Biological Chemistry

Molecular and General Genetics

Plant Cell

Plant Cell Reports

Plant Molecular Biology

Plant Physiology

Plant Physiology and Biochemistry

Plant Science

Physiologia Plantarum

Proceedings of the National Academy of Sciences USA

Science

3. GRANT APPLICATIONS

Department of Energy
National Science Foundation
United States Department of Agriculture - Agricultural
Research Service
United States Department of Agriculture - Competitive
Research Grants Program
United States-Israel Binational Science Foundation

VI. TEACHING EXPERIENCE

A. TEACHING ASSISTANT

Biochemistry Courses, University of Washington

B. FACULTY INSTRUCTOR

1. Lecture course in Plant Molecular Biology, Botany/Plant Biology 227, University of California, Davis, Spring, 1986 - 1996.
2. Laboratory course in Plant Molecular Biology, Botany/Plant Biology 228, University of California, Davis (co-instructor with Alan B. Bennett), Winter, 1990 - 1995.
3. Lecture course in Molecular Biology of Plant Development, Botany 125, University of California, Davis, Spring, 1995 - Spring, 1996.
4. Laboratory course in Plant Molecular Genetics, Plant Biology 198, University of California, Davis, Winter, 1997
5. Lecture course in Molecular and Cellular Biology of Plants, Plant Biology 113/113D, University of California, Davis, Spring, 1997 - present
6. Laboratory course in Plant Biotechnology Laboratory, Plant Biology 161B, University of California, Davis, Winter, 1999 - Winter 2000
7. Lecture course in Plant Growth and Development, Plant Biology 112/112D, University of California, Davis, Winter 2002 - present

C. COURSE FOR COMMUNITY COLLEGE AND STATE UNIVERSITY INSTRUCTORS

NSF Sponsored Workshop for the Inexperienced in Molecular Biology (Instructors: J. Christman, R. Tait, J. Harada), California State University, Sonoma, June 16-28, 1991.

VII. UNIVERSITY SERVICE

A. GRADUATE GROUP MEMBERSHIP AND AFFILIATION WITH TRAINING PROGRAMS

Biochemistry Graduate Group, 1985 - present

Biotechnology NIH Training Grant, 1990 - present

Cell and Developmental Biology Graduate Group, 1986 - present

Genetics Graduate Group, 1985 - present

Plant Biology Graduate Group (Formerly Botany and Plant Physiology Graduate Groups), 1985 - present

McKnight Foundation Fellowship Program: Biochemistry and Genetics of Plant-Pathogen Interactions, 1985 - 1989

Molecular and Cellular Biology NIH Training Grant, 1986 - present

Molecular Biology of the Plant Cell Program, 1987 - 1992

NSF-BBS Plant Cell Biology Training Grant (Co-Director with M.E. Etzler), 1990 - present

University of California Biotechnology Research and Education Program: Biotechnology-Goals for a Sustainable Agriculture: Mechanisms and Control of Plant and Animal Disease Using Recombinant DNA Techniques, 1985 - 1987

University of California Biotechnology Research and Education Program: Protein Targeting in Plants, 1988 - 1991

B. ADMINISTRATIVE CHAIR POSITIONS

1. Chair, Steering Committee for the Formation of the Section of Plant Biology, Division of Biological Sciences, 1991 - 1993
2. Co-Director, NSF Plant Cell Biology Training Program, 1990 - present.
3. Chair, Committee-In-Charge of the Plant Biology Major, 1993 - 1997
4. Chair, Graduate Program in Plant Biology, 2001 - present

VIII. LECTURES / INVITED SEMINARS

1977 Gordon Conference on Polyamines

1979 Gordon Conference on Polyamines

- 1982 American Society of Plant Physiologists Meeting, Secretary's Symposium
- 1983 Gordon Conference on Plant Molecular Biology
Cold Spring Harbor Course on Plant Molecular Biology
Ciba-Geigy Agricultural Division, Biotechnology Research, Research Triangle Park,
North Carolina
FMC Corporation, Agricultural Chemical Group, Princeton, New Jersey
University of Nebraska, Lincoln
- 1984 University of California, Davis
University of Wisconsin, Madison
Purdue University, West Lafayette, Indiana
University of California, Riverside
University of Illinois, Champaign-Urbana
Tokyo Meeting of Plant Biotechnology, Tokyo, Japan
International Symposium on Genetic Manipulation in Crops, Beijing, China
- 1985 American Society for Plant Physiologists Western Section Meeting, Plant Molecular
Genetics Symposium
- 1987 Monsanto Company, St. Louis, Missouri
University of California, Los Angeles
- 1988 Purdue University, West Lafayette, Indiana
UCLA Symposium: The Molecular Biology of Plant Development, Steamboat Springs,
Colorado (session chair and speaker)
American Society for Plant Physiologists Meeting, Gene Expression Session, Reno,
Nevada
Washington State University, Pullman
- 1989 Carnegie Institution of Washington, Stanford, California
Workshop on Embryogenic Plant Systems in Developmental Genetics and
Biotechnology, Ottawa, Ontario, Canada
Fifth Annual Crucifer Genetics Workshop, Davis, California

- American Society of Plant Physiologists Meeting, Symposium, Toronto, Ontario, Canada
- Horticultural Biotechnology Symposium (session chair), Davis, California
- Kobe University, Kobe, Japan
- Plantech Research Institute, Yokohama-City, Japan
- US-Japan Cooperative Science Program Meeting, Tokyo, Japan
- 1990 Monsanto Company, St. Louis, Missouri
- Tissue Culture Association Meeting, Houston, Texas
- US-Japan Cooperative Science Program Meeting, Davis, California
- 1991 Fourteenth Annual Symposium in Plant Physiology, Molecular Approaches to Compartmentation and Metabolic Regulation, Riverside, California
- Keystone Symposia on Molecular and Cellular Biology, The Genetic Dissection of Plant Cell Processes, Keystone, Colorado
- Twenty-seventh National Institute for Basic Biology Conference, Plant Organelle Proteins; Biosynthesis, Targeting, and Assembly, Okazaki, Japan
- Plantech Research Institute, Yokohama-City, Japan
- Nagoya University, Nagoya, Japan
- 1992 Crop Molecular Biology and Biotechnology Workshop, Banff, Alberta, Canada
- 1993 Sonoma State University, Ronhert Park, California
- XV International Botanical Congress, Tokyo, Japan (Session chair and speaker in two sessions)
- 1994 Monsanto Company, St. Louis, Missouri
- University of Wisconsin, Madison
- Fourth International Congress of Plant Molecular Biology, Amsterdam, The Netherlands
- Texas A&M University, College Station
- 1995 University of Arizona, Tucson
- Purdue University, West Lafayette, Indiana
- Jacques Monod Conference, "Signal Transduction during Plant Embryogenesis", Aussois, France

- 1996 University of California, Los Angeles
University of Alberta, Edmonton, CANADA
Calgene, Inc., Davis, California
FASEB Conference, "Plant Developmental Genetics", Saxon River, Vermont
50th Anniversary of the Korean Association of Biological Sciences, Seoul, KOREA
- 1997 Hokkaido University, JAPAN
Thirty-ninth National Institute of Basic Biology Conference, "Dynamic Aspects of Seed Maturation and Germination", Okazaki, JAPAN
Information Processing Systems in Plants Their Evolution and Function (session chair)
Davis, California
Sonoma State University, Rohnert Park, California
University of Minnesota, St. Paul, Minnesota
- 1998 Seed Physiology Meeting, Davis, California
The 7th NIAR/COE International Symposium, "Transcription Factors Controlling Plant Development", Tokyo, JAPAN
National Institute of Basic Biology Conference, Okazaki, JAPAN
Sacramento State University, California
University of Vienna Biocenter, AUSTRIA
Fifth Plant Embryogenesis Workshop, Barcelona, SPAIN
- 1999 International Botanical Congress XVI, St. Louis, Missouri
- 2000 World Congress on In Vitro Biology, San Diego, California
Eleventh International Arabidopsis Conference, Madison, Wisconsin
FASEB Summer Research Conference on Mechanisms in Plant Development, Saxon River, Vermont
The 8th International Symposium on Plant Seeds/5th Gatersleben Research Conference, Gatersleben, GERMANY
- 2001 DNA Plant Technology, Oakland, CA
University of Washington, Seattle

Society for Developmental Biology, Northwest Regional Meeting, Friday Harbor,
Washington

University of Kentucky, Lexington

IX. RESEARCH SUPPORT

A. J.J. HARADA AS PI

NSF: DCB-8518182; Developmental Regulation of Germination-Induced Genes; 4/86 - 4/89; \$202,900 (Total)

USDA: 86-CRCR-1-2172; Molecular Aspects of Disease in Higher Plants (co-PI: D. Gilchrist); 9/86 - 9/88; \$100,000 (Total)

USDA: 88-37262-3542; Molecular Analysis of Protein Targeting to Glyoxysomes; 7/88 - 7/90; \$120,000 (Total)

NSF: DCB-8819315; Gene Regulation in Glyoxysome Biogenesis; 4/89 - 4/92; \$231,460 (Total)

Plantech Research Institute; Gene Regulation during Embryogeny; 4/90 - 10/91; \$50,647 (Total)

USDA: 90-37261-5515; Peroxisome Biogenesis in Higher Plants; 9/90 - 8/93; \$171,000 (Total)

NSF: IBN-9118120; Gene Regulation in Glyoxysome Biogenesis; 4/92 - 3/94; \$200,000 (Total)

NSF: IBN-9317526; Regulation of Glyoxysome Formation during Higher Plant Development; 4/94 - 3/98; \$360,000 (Total)

DOE: DE-FG03-94ER20139; Regulation of Embryonic Development in Higher Plants; 5/94 - 5/97; \$276,069 (Total)

DOE: DE-FG03-94ER20139; Regulation of Embryonic Development in Higher Plants; J. Harada; 5/97 - 5/00; 288,000 (Total)

Ceres Inc.; Regulators of Embryo and Seed Development, 10/98 - 10/03; \$190,000 (Annual).

DOE: DE-FG03-94ER20139; Regulation of Embryonic Development in Higher Plants; J. Harada; 5/00 - 5/03; \$315,000 (Total)

B. J.J. HARADA AS CO-PI OR COOPERATOR

McKnight Foundation; Biochemistry and Genetics of Plant-Pathogen Interactions (PIs: T. Kosuge, G. Bruening, D. Gilchrist); 9/83 - 8/86 & 9/86 - 8/89; \$1,500,000 (Total)

U.C. Biotechnology Research and Training Program; Biotechnology-Goals for a Sustainable Agriculture: Mechanisms and Control of Plant and Animal Disease Using Recombinant DNA Techniques (PI: R. Micheltore); 7/85 - 6/87; \$230,000 (Total)

U.C. Biotechnology Research and Education Program; Molecular Basis of Protein Targeting to Subcellular Compartments of Plant Cells (PI: A. Bennett); 7/87 - 6/90; \$300,000 (Total)

NSF U.S.-Japan Cooperative Science Program; Plant Cell Bioprocess Engineering for Production of Secondary Metabolites (P.I.: D. Ryu); 4/89 - 3/91; \$38,000 (Total)

NSF: DIR-9014274 (Training Grant); Plant Cell Biology Training Program (Co-Directors: M. Etzler and J. Harada); 9/90 - 9/95; \$2,520,038 (Total)

NSF: DIR 8920216; Planning and Managing Center of Engineering Plants for Resistance Against Pathogens (PI: G Bruening); 2/91 - 1/96; ~\$3,500,000

NSF: BIR-9414106; Plant Cell Biology Training Program (Co-PIs: M. Etzler and J. Harada); 7/95-6/00; \$938,532

X. PUBLICATIONS

1. Coates, R.M., Conradi, R.A., Ley, D.A., Akesson, A., Harada, J., Lee, S.C., and West, C.A. (1976). Enzymatic cyclization of (R,S)-14,15-oxidogeranylgeranyl pyrophosphate to 3 α - and 3 β -hydroxykaurene. *J. Am. Chem. Soc.* 98: 4659-4661.
2. Jorstad, C.M., Harada, J.J., and Morris, D.R. (1980). Structural specificity of the spermidine requirement of an *Escherichia coli* auxotroph. *J. Bacteriol.* 141: 456-463.
3. Morris, D.R. and Harada, J.J. (1980). The participation of polyamines in the proliferation of bacterial and animal cells. In: *Polyamines in Biomedical Research*, J. Gaugas, ed., John Wiley and Sons, Ltd., London, pp. 1-16.
4. Harada, J.J. (1981). Effects of polyamine limitation on the growth of animal cells. Ph.D. Thesis, University of Washington, Seattle, Washington.
5. Harada, J.J., Porter, C.W., and Morris, D.R. (1981). Induction of polyamine limitation in Chinese hamster ovary cells by α -methyl-ornithine. *J. Cell. Physiol.* 107: 413-426.
6. Harada, J.J. and Morris, D.R. (1981). Cell cycle parameters of Chinese hamster ovary cells during exponential, polyamine-limited growth. *Molec. Cell. Biol.* 1: 594-599.
7. Young, E.T., Menard, R.C., and Harada, J. (1981). Monocistronic and polycistronic bacteriophage T4 gene 23 messages. *J. Virol.* 40: 790-799.
8. Goldberg, R.B., Fischer, R.B., Harada, J.J., Jofuku, D., and Okamuro, J.K. (1983). Organization of soybean seed protein genes and their flanking regions. In: *Structure and Function of Plant Genomes*, O. Ciferri and L. Dure III, eds., Plenum Publishing Corporation, New York, pp. 37-45.

9. Bruening, G., Harada, J., Kosuge, T., and Hollaender, A. eds. (1987). *Tailoring Genes for Crop Improvement: an Agricultural Perspective*, Plenum Press, New York.
10. Barker, S.J., Harada, J.J., and Goldberg, R.B. (1988). Cellular localization of soybean storage protein mRNA in transformed tobacco seeds. *Proc. Natl. Acad. Sci. USA* 85: 458-462.
11. Harada, J.J., Baden, C.S., and Comai, L. (1988). Spatially regulated genes expressed during seed germination and postgerminative development are activated during embryogeny. *Mol. Gen. Genet.* 212: 466-473.
12. Harada, J.J., Dietrich, R.A., Comai, L., and Baden, C.S. (1988). Regulation of gene expression during seed germination and postgerminative development. In: *Plant Gene Research, Volume 5, Temporal and Spatial Regulation of Plant Genes*, D.P.S. Verma and R.B. Goldberg, eds., Springer-Verlag, New York, pp. 26-39.
13. Dietrich, R.A., Maslyar, D.J., Heupel, R.C., and Harada, J.J. (1989). Spatial patterns of gene expression in *Brassica napus* seedlings: Identification of a cortex-specific gene and localization of mRNAs encoding isocitrate lyase and a polypeptide homologous to proteinases. *Plant Cell* 1: 73-80.
14. Comai, L., Baden, C.S., and Harada, J.J. (1989). Deduced sequence of a malate synthase polypeptide encoded by a subclass of the gene family. *J. Biol. Chem.* 264: 2778-2782.
15. Comai, L., Dietrich, R.A., Maslyar, D.J., Baden, C.S., and Harada, J.J. (1989). Coordinate expression of transcriptionally regulated isocitrate lyase and malate synthase genes in *Brassica napus* L. *Plant Cell* 1: 293-300.
16. Harada, J.J., DeLisle, A.J., Baden, C.S., and Crouch, M.L. (1989). Unusual sequence of an abscisic acid-inducible mRNA which accumulates late in *Brassica napus* seed development. *Plant Mol. Biol.* 12: 395-401.
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18. Gilchrist, D.G. and Harada, J.J. (1989). Mode and physiological consequence of AAL-toxin interaction with the *asc* locus in tomato. In: *Phytotoxins and Plant Pathogenesis*, NATO ASI Series, Vol. H27, A. Graniti et al., eds., Springer-Verlag, Berlin, pp. 113-121.
19. Dure, L. III., Crouch, M., Harada, J., Ho, T.-H.D., Mundy, J., Quatrano, R., Thomas, T., and Sung, Z.R. (1989). Common amino acid sequence domains among the LEA proteins of higher plants. *Plant Mol. Biol.* 12: 475-486.
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22. Ettinger, W.F. and Harada, J.J. (1990). Translational or posttranslational processes affect differentially the accumulation of isocitrate lyase and malate synthase proteins and enzyme activities in embryos and seedlings of *Brassica napus*. Arch. Biochem. Biophys. 281: 139-143.
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24. Harada, J.J. (1990). Summary: Biotic Stress. In: Horticultural Biotechnology, A.B. Bennett and S.D. O'Neill, eds., Wiley-Liss, Inc., New York, pp. 261-262.
25. Olsen, L.J. and Harada, J.J. (1991). Biogenesis of peroxisomes in higher plants. In: *Molecular Approaches to Compartmentation and Metabolic Regulation*, A.H.C. Huang and Lincoln Taiz, eds., The American Society of Plant Physiologists, Rockville, MD, pp. 129-137.
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27. Comai, L., Matsudaira, K.L., Heupel, R.C., Dietrich, R.A., and Harada, J.J. (1992). Expression of a *Brassica napus* malate synthase gene in transgenic tomato plants during the transition from late embryogeny to germination. Plant Physiol. 98: 53-61.
28. Dietrich, R.A., Radke, S.E., and Harada, J.J. (1992). Downstream DNA sequences are required to activate a gene expressed in the root cortex of embryos and seedlings. Plant Cell 4: 1371-1382.
29. Zhang, J.Z., Gomez-Pedrozo, M., Baden, C.S., and Harada, J.J. (1993). Two classes of isocitrate lyase genes are expressed during late embryogeny and postgermination in *Brassica napus* L. Mol. Gen. Genet. 238: 177-184.
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32. Zhang, J.Z., Laudencia-Chingcuanco, D.L., Comai, L., Li, M., and Harada, J.J. (1994). Isocitrate lyase and malate synthase genes from *Brassica napus* are active in pollen. Plant Physiol. 104: 857-864.
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36. Olsen, L.J. and Harada, J.J. (1995). Peroxisomes and their assembly in higher plants. *Annu. Rev. Plant Physiol. Plant Mol. Biol.* 46: 123-146.
37. Zhang, J.Z., Santes, C.M., Engel, M.L., Gasser, C.S., and Harada, J.J. (1996). DNA sequences that activate isocitrate lyase gene expression during late embryogenesis and during postgerminative growth. *Plant Physiol.* 110: 1069-1079.
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43. Harada, J.J., Lotan, T., Fischer, R.L., and Goldberg, R.B. (1998). ...response: Embryos without sex. *Trends Plant Sci.* 3: 452-453.
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45. Ohad, N., Yadegari, R., Margossian, L., Hannon, M., Michaeli, D., Harada, J.J., Goldberg, R.B., and Fischer, R.L. (1999). Mutations in *FIE*, a WD polycomb group gene, allow endosperm development without fertilization. *Plant Cell* 11: 407-416.
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49. Harada, J.J. (2001). Role of Arabidopsis LEAFY COTYLEDON genes in seed development. *J. Plant Phys.* 158:405-409.
50. Apuya, N. R., Yadegari, R. Fischer, R.L., Harada, J.J. Zimmerman, J.L., and Goldberg, R.B. (2001). The Arabidopsis embryo mutant *schlepperless* has a defect in the chaperonin-60alpha gene. *Plant Phys.* 126:717-730.
51. Stone, S.L., Kwong, L.W., Matsudaira Yee, K., Pelletier, J., Lepiniec, L., Fischer, R.L., Goldberg, R.B., and Harada, John J. (2001). *LEAFY COTYLEDON2* encodes a B3 domain transcription factor that induces embryo development. *Proc. Nat. Acad. Sci. USA*, 98: 11806-11811.
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53. Weterings, K., Apuya, N.R., Bi, Y., Fischer, R.L., Harada, J.J., and Goldberg, R.B. (2001). Regional localization of suspensor mRNAs during early embryo development. *Plant Cell*. 13:2409-2425.

XI. PATENTS

1. Harada, J.J. and Zhang, J.Z. Plant promoter sequences useful for gene expression in seeds and seedlings. U.S. Patent No. 5,689,040
2. Fischer, R.L., Ohad, N., Kiyosue, T., Yadegari, R., Margossian, L., Harada, J., and Goldberg, R.B., Nucleic acids that control endosperm development in plants, US Patent No. 6,229,064
3. Harada, J.J., Lotan, T., Ohto, M., Goldberg, R.B., and Fischer, R.L., LEAFY COTYLEDON1 genes and methods of modulating embryo development in transgenic plants. US Patent No. 6,235,975

XII. STUDENTS AND POSTDOCTORAL ASSOCIATES TRAINED

A. POSTDOCTORAL ASSOCIATES

Dr. Barney Ward, 10/85-6/88; current position, Research Associate, Department of Plant Pathology, University of California, Davis

Dr. William F. Ettinger, 10/87-2/90; current position, Associate Professor, Department of Biology, Gonzaga University, Spokane, WA

Dr. Mariza Gomez-Pedroso, 5/88-10/88; current position, unknown

Dr. Edward J. Newbigin, 2/89-3/90 (Joint postdoc with M.E. Etzler); current position, Laboratory Director, School of Botany, University of Melbourne

Dr. Laura J. Olsen, 4/90-8/93; current position, Associate Professor, Department of Biology, University of Michigan

Dr. Marilyn A.L. West, 1/92-4/95; current position, Lab Manager, Department of Vegetable Crops, University of California, Davis

Dr. Christina Santes Valera, 1/94-12/96; current position, science teacher, Davis, California

Dr. Tamar Lotan, 4/95-7/98; current position, establishing biotechnology company in Israel

Dr. Kazutoshi Yamagishi, 4/95-5/98; current position, Postdoctoral Fellow, RIKEN, Japan

Dr. Masa-aki Ohto, 5/95-2/96 & 5/01-present; past position, Assistant Professor, Department of Plant Biochemistry, National Institute of Basic Biology, Okazaki, Japan

Dr. Luis Perez Grau, 3/00-6/01; current position, Scientist, Simplot Corporation, Boise, ID

Dr. Sandra L. Stone, 5/01-present

B. GRADUATE STUDENTS

Lucio Comai, Ph.D., Biochemistry, December, 1990. Thesis: Regulation of malate synthase gene expression during embryogeny and postgermination in *Brassica napus*. Damon-Runyon Postdoctoral Fellow with R. Tjian, University of California, Berkeley. Current position, Associate Professor, Department of Microbiology, University of Southern California

Robert A. Dietrich, Ph.D. Genetics, September, 1991, McKnight Graduate Fellow. Thesis: Regulation of a *Brassica napus* gene expressed during embryogenesis and postgerminative growth. Current Position: Scientist, Syngentia, Inc., Research Triangle

Park, NC

James Z. Zhang, Ph.D. Genetics, March, 1993. Thesis: Developmental Regulation of Isocitrate Lyase Gene Expression in *Brassica napus* L. Current Position: Scientist, Mendel Biotechnology, Hayward, CA

Debbie L. Laudencia-Chingcuanco, Ph.D. Genetics, December 1994, UC Dissertation Fellow. Thesis: Functional Analysis of the Regulatory DNA Sequences of a Malate Synthase Gene from *Brassica napus*. Current position, NSF Postdoctoral Fellow with S. Hake, University of California, Berkeley.

Elizabeth A. Wasson, M.S. Plant Biology, July, 1994, Plant Cell Biology Graduate Fellow.

Martin P. Doyle, M.S. Plant Biology, August, 1995, Plant Cell Biology Graduate Fellow.

Minsung Kim, October, 1996 - 1998, Genetics Graduate Group Ph.D. student.

Raymond W. Kwong, September, 1998 - present, Plant Biology Graduate Group Ph.D. Student

Hye-seung Lee, September, 1998 - present, Plant Biology Graduate Group Ph.D. Student

C. **UNDERGRADUATE STUDENTS (NAME, YEARS INVOLVED IN RESEARCH, CURRENT POSITION)**

Davila, Alvaro, 1985, Unknown

Goldman, Polly, 1986, Graduate student, UCSC

Maslyar, Daniel, 1987 - 1990, Medical student, UCSF

Nguyenle, Thuylinh, 1989 - 1990, Unknown

Holst, Eric, 1990, Graduate student, Duke University

Morales, Consuelo, 1990 - 1995, Employed

Danao, Jay, 1990 - 1995, Employed

Li, Morris, 1991, Medical student, Loyola University

Fernandez, Lewis, 1991, Graduate school

Chan, Victor, 1991 - 1992, Technician, Sandoz

Pham, Nhu, 1991, Unknown

Hartt, Gregory, 1992 - 1993, M.D./Ph.D. student, Ohio State University

Tran, Minh, 1992, Pharmacy student, Creighton University

Willard, Traci, 1992, Employed

Renouf, Lynette, 1992, Unknown

Culberson, Carla, 1992, Unknown

Hague, Carolyn, 1993, Employed

Engel, Michelle, 1993 - 1994, Graduate student, UCLA

Togioka, Patti, 1993 - 1994, Employed

Dung, Hoa, 1993 - 1994, Employed

Lo, Russell, 1993 - 1995, Ph.D. student, University of Washington

Heraux, Jonathan, 1994, Student, University of Hawaii

Gabaldon, Amapro, 1994, Student, University of Madrid

Anthony Phengrasamy, 1995, Pharmacy student, UCSF
Wortley, Sara, 1995 - 1997, unknown
Swirsding, Kendra 1995 - 1997, Technician
Leong, Mary , 1995 - 1997, Optometry School
Chao, Kim 1995- 1997, Unknown
Lee, Vong 1995 - 1996, Unknown
Baxter, Kim, 1996 - 1997, Graduate Student
Kwong, Raymond, 1996 - 1997, Graduate Student, UCD
Jennifer Lee, 1997, 1998 - 1999, Medical Student, UCSF
Linda Kwong, 1997- 1999, Technician
Michael Gonzales, 1997 - 1999, Graduate Student, University of Wisconsin, Madison
Yue-yun To, 1998, unknown
Tereza Kolesnikov, 1998 - 1999, Technician, Stanford University
Kyle Mizuno, 1998 - 1999, Biochemist
Diana Lee, 1999 - 2000, Student
Christy Ferlatte, 2000-2001, Student
Stephanie Paula, 2001-present, Student
Abeba Kiros, 2001-present, Student